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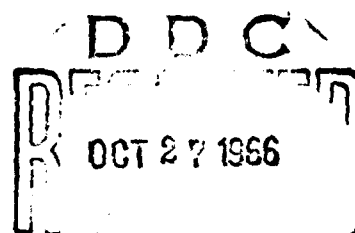
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Streptomycin Inhibition of Elaboration of Staphylococcal Enterotoxigenic Protein

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Staphylococcus aureus strain S-6, under certain cultural conditions, synthesizes and elaborates into the medium an enterotoxigenic protein (M. S. Bergdoll, M. J. Surgalla, and G. M. Dack, J. Immunol. 83:334, 1959). We have observed that streptomycin interferes with the elaboration of this protein by a streptomycin-resistant isolate of strain S-6 at drug levels permitting continued synthesis of those proteins necessary for growth and multiplication of the organisms. Enterotoxigenic

protein-sensitive isolate). The cultures were shaken [100 3-inch (7.6-cm) strokes per minute] at 37 C for 24 hr, after which the optical density

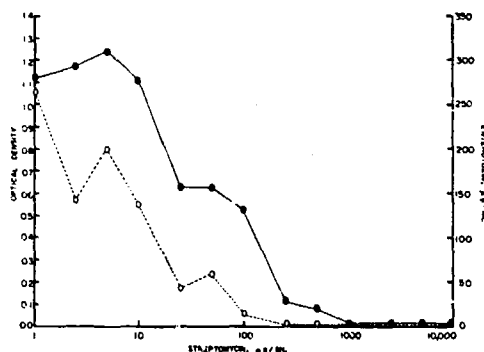


FIG. 1. Growth and synthesis of enterotoxin by streptomycin-sensitive *Staphylococcus aureus* in presence of streptomycin. Symbols: ●, optical density; ○, enterotoxin.

production in the streptomycin-dependent strain, even at the point of maximal population increase, was severely depressed.

Streptomycin sulfate (E. R. Squibb & Sons, New York, N.Y.) was added at various levels to a medium composed of N-Z-Amine type A (Sheffield Chemical, Norwich, N.Y.), 4.0%; yeast extract (Difco), 0.4%; K_2HPO_4 , 0.1%; and tap water, at pH 6.8 to 7.0. A 25-ml amount of this medium in a 250-ml Erlenmeyer flask was inoculated (1.0%, v/v) with a culture of a phage-free isolate of *S. aureus* S-6. Isolates sensitive, resistant, and dependent with regard to streptomycin were used (the resistant and dependent isolates were derived as one-step mutants from the strep-

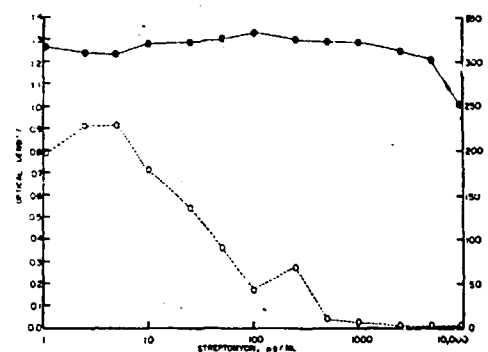


FIG. 2. Growth and synthesis of enterotoxin by streptomycin-resistant *Staphylococcus aureus* in presence of streptomycin. Symbols: ●, optical density; ○, enterotoxin.

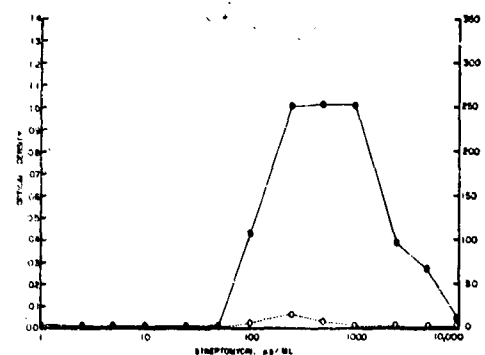


FIG. 3. Growth and synthesis of enterotoxin by streptomycin-dependent *Staphylococcus aureus* in presence of streptomycin. Symbols: ●, optical density; ○, enterotoxin.

(1:10 dilution) and enterotoxin level were determined, the latter by the Oudin (Methods Med. Res. 5:335, 1952) method with the use of specific antiserum in an agar-gel diffusion system.

Figure 1 shows that the enterotoxin level declined with optical density when the streptomycin-sensitive strain was treated with increasing concentrations of streptomycin. With the resistant strain of the organism (Fig. 2), the enterotoxin level was decreased over a broad range of drug concentration, in which there was no inhibition of culture growth. Although streptomycin at 250 to 1,000 $\mu\text{g/ml}$ was sufficient to allow normal culture growth of the streptomycin-dependent strain (Fig. 3), the maximal detectable enterotoxin protein was less than 10% of that obtained with the sensitive or resistant strains, and occurred at the lowest level of drug permitting normal culture growth.

These data show that streptomycin limits the

elaboration of immunologically detectable enterotoxin in both resistant and dependent organisms under conditions in which they apparently are functioning normally in other respects. The reasons for this are not clear. Hypotheses based on the similarity of sites of protein synthesis and those of streptomycin action are tempting. Because enterotoxin is not detectable until it reaches the cell surface (M. E. Friedman and J. D. White, *J. Bacteriol.* 89:1155, 1965), the cell membrane would be of particular interest. It is unlikely, under current concepts, that the protein suffers from miscoding. It is more likely that it is incomplete, or that it is not released from the cell because of streptomycin-induced alterations in the cell itself.